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PRINCIPAL INVESTIGATOR:

Vernon Lin, MD PhD

CONTRACTING ORGANIZATION: Cleveland Clinic Foundation
Cleveland, Ohio

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14. ABSTRACT This award has 4 separate studies. Study 1: The penicillin-induced seizure animal model was generated by acute focal intracortical injection of penicillin in the motor cortex of rats. The effects of functional magnetic stimulation (FMS) on penicillin-induced seizure were investigated using behavioral recording and electroencephalographic (EEG) recording and results were published. Study 2: The motor cortex (M1) and the corticospinal tracts (CST) were directly modulated using brain stimulation to benefit rehabilitative outcomes of upper limb training in incomplete SCI (iSCI). Eight patients and three healthy control subjects were enrolled. SCI patients receiving brain stimulation showed greater benefit in upper limb function than those receiving rehabilitation alone. Study 3: The efficacy of using FMS for respiratory muscle conditioning was evaluated in patients with multiple sclerosis (MS). Four patients with MS were enrolled. A 6-week FMS conditioning of the expiratory muscles improved voluntary expiratory functions, indicating that FMS may be a potential noninvasive therapeutic technology for training respiratory muscles in patients with MS. Study 4: This pilot study will evaluate the usefulness of FMS as a noninvasive method to stimulate the GI motility in individuals with non-neurological constipation. The PI worked with HRPO to modify the protocol (remove the sham treatment).					
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Introduction

Over the years the Functional Magnetic Stimulation (FMS) Laboratory has focused on studying the effectiveness of applying FMS for improving functions in patients with spinal cord injury and spinal cord disorders (SCI/D). Using animals and humans we have successfully demonstrated that FMS is effective for improving cough, bladder function, gastric emptying and colonic transits in SCI/D. This proposal consists of four pilot projects with major aims to determine the mechanisms behind some novel applications of FMS, such as for epilepsy control (study 1), and for improving upper limb function in SCI/D (study 2); and to explore additional clinical benefits of FMS, such as for improving constipation in general population (study 4), and for improving cough in patients with multiple sclerosis (study 3). The following paragraphs provide brief synopsis of these four pilot projects.

Study 1: Magnetic stimulation and epilepsy

In this study, transcranial magnetic stimulation (TMS) will be tested for its efficacy in controlling seizure in a rat seizure model. In addition, a cell culture system will be used to test the mechanisms of the effects of magnetic stimulation on axonal outgrowth and the hypothesis that these responses are mediated by brain-derived neurotrophic factor (BDNF).

Study 2: Directing Neuroplasticity to Improve Rehabilitative Outcomes of the Upper Limb in Incomplete Quadriplegia

In this study, the motor cortex (M1) and the corticospinal tracts (CST) will be directly modulated using brain stimulation to benefit rehabilitative outcomes of upper limb training in incomplete SCI (iSCI). Functional and structural mechanisms of such plasticity, we expect, will be demonstrable using Transcranial Magnetic Stimulation (TMS) and Diffusion Tensor Imaging (DTI) in patients.

Study 3: Expiratory Muscle Conditioning Using Functional Magnetic Stimulation for Patients with Multiple Sclerosis

In this study, we will investigate the efficacy of using FMS technique for respiratory muscle conditioning in patients with multiple sclerosis; and will compare the results of the expired functions (volume, pressure, and flow) generated by using the FMS technique with data obtained from using the resistive expiratory muscle training (REMT) methodology.

Study 4: A prospective trial to study whether functional magnetic stimulation enhances gastrointestinal motility in patients with chronic constipation

This pilot study will evaluate the usefulness of FMS as a noninvasive method to stimulate the GI motility in individuals with non-neurological constipation by adopting a 5-week conditioning protocol. This program may demonstrate that FMS can be an effective treatment modality for patients with constipation and thus promote health, independence, and quality of life.

Annual Progress Report

For the period of September 15, 2013 through September 30, 2014

I. BASIC INFORMATION

SUBJECT: Magnetic stimulation and epilepsy

AWARD #: W81XWH-11-1-0707

CCF IACUC#: 2010-0415 (expires March 6, 2014)

SPONSOR: Department of Defense (DoD) Telemedicine and Advanced
Technology Research Center

PRINCIPAL INVESTIGATOR: Ching-Yi Lin PhD

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REPORT DATE : November 07, 2014

II. STUDY PROGRESS

A. Introduction: Study Progress in Relation to Investigational Plan

During previous 22 months of project, we have reached several milestones according to the time frame that has been proposed in the grant. For the Specific Aim 3, the penicillin-induced seizure animal model has been generated by acute focal intracortical injection of penicillin in the motor cortex of rats. The effects of transcranial magnetic stimulation (TMS) on penicillin-induced seizure have been investigated using behavioral recording and electroencephalographic (EEG) recording. The results obtained have been published to *Brain Research* (2014).

For the Specific Aim 1, the effects of MS on the level of BDNF; (ii) the roles of BDNF on the MS regulation of neurite outgrowth have been studied. The results have been submitted to *Journal of Neural Engineering*.

To test the hypotheses that MS-regulated neuronal activity can be translated to specific changes in neuronal arborization and thus regulate synaptic activity and function, we examine the effects of MS on neurite growth by Neuroscreen-1 (NS-1) cells over the pulse frequencies 1, 5 and 10 Hz. Plated NS-1 cells were exposed to MS twice per day for 3 days and then evaluated for length and number of neurites. We have demonstrated that MS at both 30% and 40% machine output (MO) dramatically affect neurite growth on growing axons but not on dendrites. MS effects were frequency-dependent, most evident in the bolstering of axonal outgrowth only seen at 10 Hz MS group. Using gridded coverslips in our MS protocol enabled us to distinguish two electromagnetic responses, where the center region of the coverslip received minimal MS-induced current density (zone 1) and the remaining area experienced maximal MS-induced current density (zone 2). We have demonstrated that MS-increased axonal growth was most evident in zone 1, but not in zone 2. Furthermore, we found that MS increased brain-derived neurotrophic factor (BDNF) expression and secretion in a frequency-dependent manner. Taken together, our results show that MS exerts distinct effects when different frequencies and intensities are applied on the neuritic compartments (axon versus dendrite) of NS-1 cells. These findings support the concept that MS increases BDNF expression and signaling, which sculpts axonal arborization and connectivity by which neuronal activity is regulated. Understanding the mechanism behind MS action is crucial in order to efficiently incorporate its use into potential therapeutic strategies.

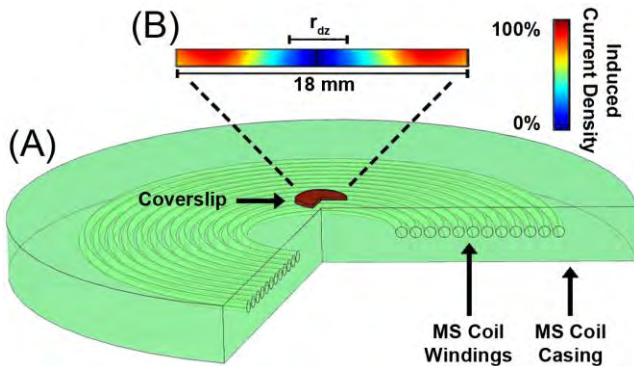


Figure 1. Model of experimental setup and MS-induced current density of cultured NS-1 cells. (A) A 3-D model of the experimental setup was constructed using the COMSOL Multiphysics® CAD software, including the geometries and relative positions of both MS coil and coverslip during stimulations. Defined by the electric current per unit area of a cross section, electric current density was estimated in the COMSOL® simulation in conjunction with fundamental electromagnetic

induction laws, Maxwell-Faraday Equations. (B) Due to the nature of circular MS coils, negligible current is induced from the center of the MS coil, described as the dead zone radius (r_{dz}). The color bar represents the induced current density spread in the coverslip in A/mm², where the maximum (~35 mA) and minimum (within the dead zone radius) are colored red and blue, respectively.

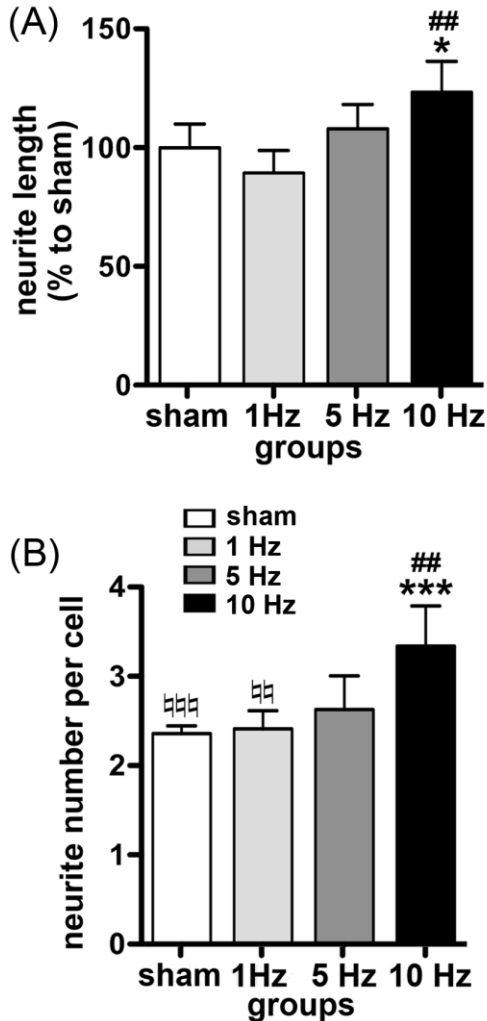


Figure 2. Neurite length and number were regulated by MS. NS-1 cells were cultured onto a collagen-coated coverslip. MS (1, 5 or 10 Hz) at 30% MO was applied starting from the second day, twice per day, for the next 3 days. The NS-1 cells were then fixed and stained for HCS CellMask Red. (A) NS-1 cells extended longer neurites, as compared to the sham group, after 3 days of treatment with 10 Hz MS, but not 1 Hz or 5 Hz MS. (B) The average number of neurites per NS-1 cell was significantly increased by 10 Hz MS. Bars show mean \pm SEM values. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.005$ vs. sham MS group; # $p < 0.05$, ## $p < 0.01$, ### $p < 0.005$ vs. 1 Hz MS group; $\Omega\Omega$ $p < 0.01$, $\Omega\Omega\Omega$ $p < 0.005$ vs. 5 Hz MS group; \natural $p < 0.05$, $\natural\natural$ $p < 0.01$, $\natural\natural\natural$ $p < 0.005$ vs. 10 Hz MS group; Two-way ANOVA with Bonferroni *post-hoc* tests.

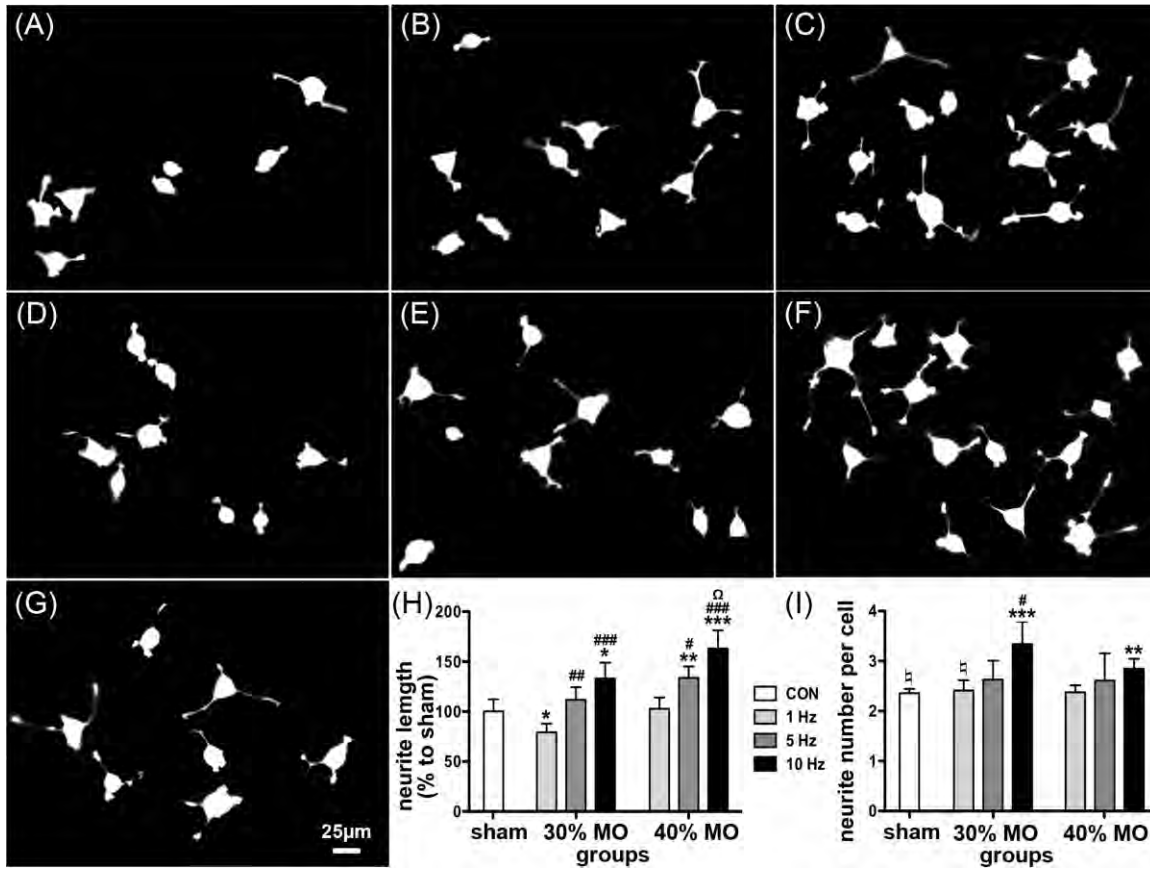


Figure 3. Neurite growth was regulated by both MS frequency and intensity. Representative images show that neurite lengths and counts per NS-1 cell were regulated by MS, as compared to the sham MS group (G). NS-1 cells treated with 1 Hz MS (A, D) expressed shorter neurites, whereas 5 Hz MS (B, E) had negligible effects. 10 Hz MS (C, F) however, consistently expressed longer neurites and higher counts per cell. The graphs illustrate average neurite lengths (H) and neurite counts (I) for NS-1 cells treated with sham MS (sham) and all MS treatment groups (1, 5, or 10 Hz) at 30% MO or 40% MO. Bars showed mean \pm SEM values. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.005$ vs. sham MS group; # $p < 0.05$, ## $p < 0.01$ vs. 1 Hz MS group; ‡ $p < 0.05$ vs. 10 Hz MS group; Two-way ANOVA with Bonferroni *post-hoc* tests.

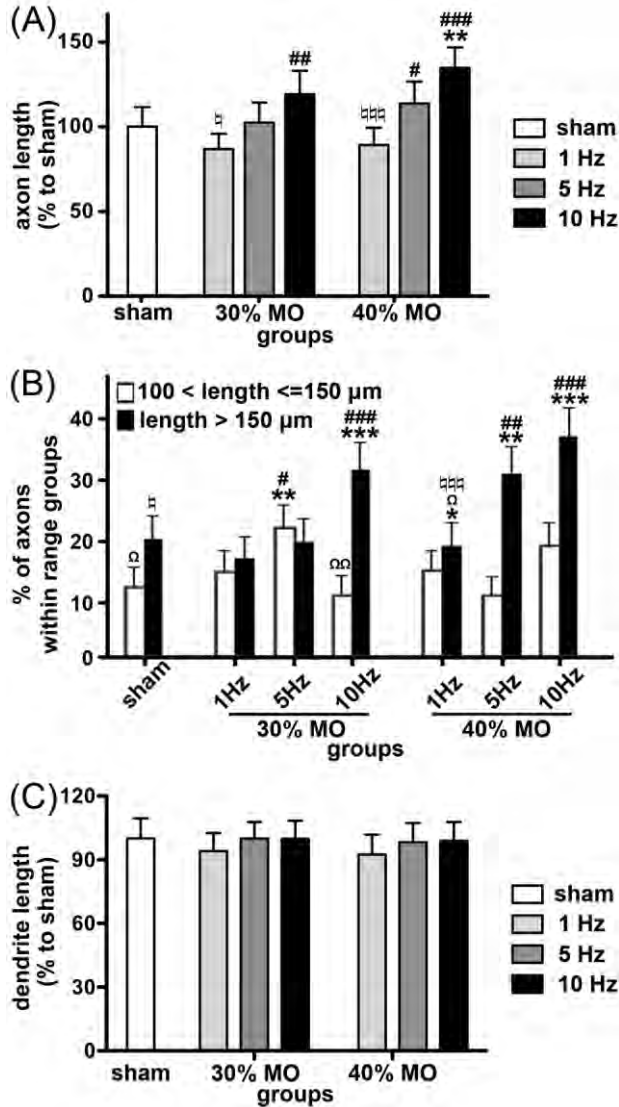


Figure 4. Degree of MS influence over axon and dendrite lengths. Neurite outgrowth measurements were broken down into axons and dendrites for further analysis. Graphs show either axon (A) or dendrite (C) lengths measured for NS-1 cells fixed with sham MS treatment (sham) or MS (1, 5 or 10 Hz) at 30% or 40% MO. (B) further illustrates any trends in the percentage of axons with specified greater length ranges for all test groups. Bars show mean \pm SEM values. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.005$ vs. sham MS group; # $p < 0.05$, ## $p < 0.01$, ### $p < 0.005$ vs. 1 Hz MS group; Ω $p < 0.05$, $\Omega\Omega$ $p < 0.01$ vs. 5 Hz MS group; \S $p < 0.05$, $\S\S\S$ $p < 0.005$ vs. 10 Hz MS group; Two-way ANOVA with Bonferroni *post-hoc* tests.

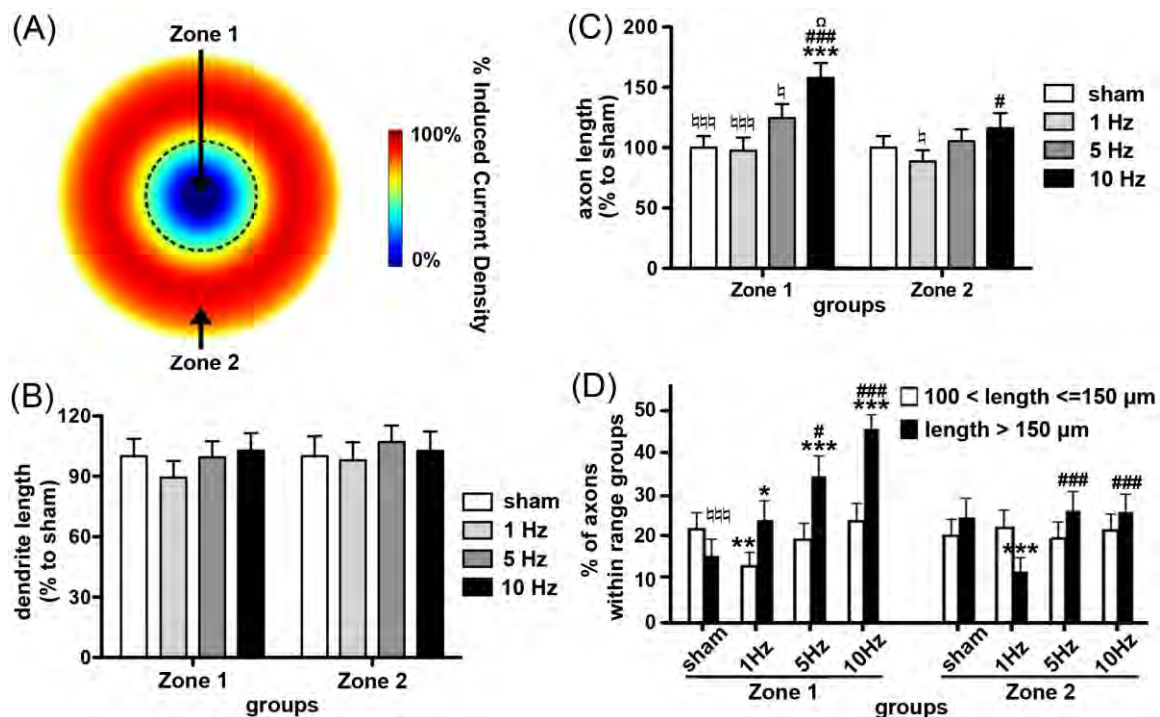


Figure 5. MS intensity differentially increased axonal growth via double-zone analysis. The following results were derived from three representative experiments measuring neurite length and number in zones 1 and 2 of gridded coverslips after MS (30% MO) to either 1, 5, or 10 Hz at 3-d. (A) Schematic of the gridded coverslip divided into zone 1 and zone 2, where NS-1 cells received minimal (zone 1) and maximal (zone 2) MS-induced current densities, respectively. The gridded coverslips were categorized under zones 1 and 2 to distinguish regions either in or out r_{dz} and their respective electric field distribution in our experimental model. If we consider 100% induced current density as maximal MS efficiency, zone 1 NS-1 cells were subjected to <10% efficiency, whereas zone 2 was primarily subject to ~90% efficiency. Zone differential data are illustrated via dendrite (B) and axon (C) lengths and percentage of axons within specified greater lengths range groups (D). Bars show mean \pm SEM values. * $p<0.05$, ** $p<0.01$, *** $p<0.005$ vs. sham MS group; # $p<0.05$, ### $p<0.005$ vs. 1 Hz MS group; Ω $p<0.05$, ΩΩ $p<0.01$ vs. 5 Hz MS group; ‡ $p<0.05$, ‡‡‡ $p<0.005$ vs. 10 Hz MS group; Two-way ANOVA with Bonferroni *post-hoc* tests.

B. Investigators/Investigational Sites

The study is being conducted at only one site – Cleveland Clinic Foundation. There are no plans to add another site.

Investigational Site	Investigators
Cleveland Clinic Foundation 9500 Euclid Avenue, Cleveland, OH 44195	Ching-Yi Lin PhD

C. Number of Subjects / Animals

N/A

D. Summary of Anticipated and Unanticipated Adverse Effects

N/A

E. Investigational Devices

N/A

F. Deviations from Investigational Plan

No deviations from the investigational plan.

III. RISK ANALYSIS

A. New Adverse Information and New Risk Analysis

No new risks have been identified that would require a new risk analysis.

IV. OTHER CHANGES

N/A

V. FUTURE PLANS

In order to move our research findings forward to clinical application, we have started to investigate the changes in brain oscillation after seizure induction/prog and/or magnetic stimulation.

Annual Progress Report

For the period of October 1, 2013 through September 30, 2014

I. BASIC INFORMATION

AWARD NUMBER: W81XWH-11-10707

IRB NUMBER: 11-823 (expiration date: 9/8/2015)

STUDY NAME: Directing Neuroplasticity to Improve Rehabilitative Outcomes of the Upper Limb in Incomplete Quadriplegia

SPONSOR: Department of Defense (DoD) Telemedicine and Advance Technology Research Center

PI: Ela Plow PhD PT
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REPORT DATE : November 7, 2014

I. STUDY PROGRESS

A. Summary: Study Progress in Relation to Investigational Plan

The long-term objective of our study is to maximize the rehabilitative potential in spinal cord injury (SCI). We are addressing this goal by harnessing the maximal potential available for neuroplasticity in patients with SCI. SCI is an important cause of serious, long-term disability in young adults. Upper limb dysfunction is one of the most prevalent and debilitating impairments. More than 75% of patients with quadriplegia (weakness/paralysis of all 4 limbs) prioritize return of upper limb function over any other lost function.

Although various therapeutic programs have been employed to mitigate functional impairments of the arm and hand, effects are weak and invariable. *Limited success of rehabilitation is speculated to be associated with maladaptive changes in the brain*; for instance, maps in the motor cortex (M1) devoted to less-affected muscles of weak limbs expand at the expense of maps devoted to the more affected muscles, limiting effects of training of the more affected segments. Recovery can be served by adaptive changes of maps in the M1 that potentially improve descending motor output to affected limbs.

Our **objective** has been to directly modulate plasticity in M1 using brain stimulation. Our **central hypothesis** is that noninvasive brain stimulation, called transcranial direct current stimulation (tDCS), when directed to maps of affected upper limb in M1 will generate functional advantage for rehabilitation. Thus, patients are assigned randomly to either the tDCS plus rehabilitation group or the sham plus rehabilitation group. We premise that rehabilitative outcomes of paralyzed upper limbs will improve in patients in both groups, but they will improve more significantly for patients in the tDCS plus rehabilitation group. The underlying mechanism would involve reorganization of maps; maps of affected parts would expand at the expense of those of less affected, as demonstrated with noninvasive Transcranial Magnetic Stimulation (TMS) and an MRI method called Diffusion Tensor Imaging (DTI).

Towards our objective, we have now enrolled 8 patients and 3 healthy control subjects. Last two patients are completing interventions of the study and with time for follow-up will be completing the study procedures by Sept 2015. This is tremendous progress. We have already met >80% of the anticipated number of patients. Our results amongs patients who have completed the study after enrollment are listed here.

B. Study progress and request for no-cost extension

The progress of the study was challenging at first. We have now submitted a request for approval to continue use of remaining funds through a no-cost extension period based on the following factors.

1. Despite delayed start, and slow enrollment, our recruitment efforts have been successful, we have enrolled > 80% of the anticipated number of patients already and additional <20% patients will be recruited by Nov 2014. Since we are now benefitting from enhanced recruitment, we are seeking approval to complete study procedures on the already enrolled patients who are now completing therapies or are soon due to begin therapies. For instance, patients who start this month and next would require 1.5 months each to complete study therapies and then will be required to undergo one additional follow-up visit 3 months later.

The reason for late start despite steady enrollment is that scheduling travel and lodging for a lot of these patients with paralysis following spinal cord injury poses the biggest challenge. Some of them are from out-of-town and can only manage studies in summer. We believe by extending the study by 1 more year, we would be able to complete recruitment goals and collect all data (including their 3-month follow-up visit) on all enrollees. This we believe would not impose any more costs on the budget than what remains for patients' care.

Therefore, since the study recruitment and data collection processes are proceeding well, we have requested extension to simply collect all procedures on currently enrolled (>80% of anticipated) and due-to-be enrolled (<20% of anticipated) patients.

2. While the enrollment has been challenging, **we have made tremendous intellectual progress on study**, which although difficult initially is now rewarding
 - We have been the **first to optimize combination of brain mapping with TMS with DTI MRI** that is funded through this study. Based on these efforts, we have been awarded another federal grant (**American Heart Association**) as we have been the first to develop the approach. This approach was extensively challenging and we were able to complete only through ability to collect data on SCI patient and controls on this study.
 - We are being recognized amongst peers for our work in spinal cord injury rehabilitation and have been invited to contribute an editorial for one of the most prestigious journals, '**Neurology**' pertaining to our approach.
 - The study is advertised as a national clinical trial
 - We have been invited to publish regarding spinal cord injury work in Frontiers in Rehabilitation and present at the **meeting of the American Spinal Cord Injury Professionals**.
 - **We were recently invited to submit and have successfully submitted a grant application (in eth amount of \$2,522,000) for the Department of Defense's, Congressionally Directed Medical Research Programs, called the "Spinal Cord Injury Research Program Clinical Trial Award for Funding Opportunity Number: W81XWH-14-SCIRP-CTA". This application is a direct extension of the present DoD award.**
 - Based on our clinical trial findings, we are now submitting manuscripts for publication.

Therefore, although initial start of study was delayed due to human subject approval issues, and recruitment was challenging, we have made tremendous progress and are nearing completion of all study related procedures in remaining year of award. We have been successful not only in publishing via this award, but also in receiving grants and in being selected and invited for larger federally-funded grant applications.

B. Investigators/Investigational Sites

The study is being conducted at only one site – Cleveland Clinic Foundation. There are no plans to add another site.

Investigational Site	Investigators
Cleveland Clinic Foundation 9500 Euclid Avenue, Cleveland, OH 44195	Ela Plow PhD PT Frederick Frost MD Ken Sakaie PhD

C. Number of Subjects

Eight SCI patients and three control subjects have been enrolled.

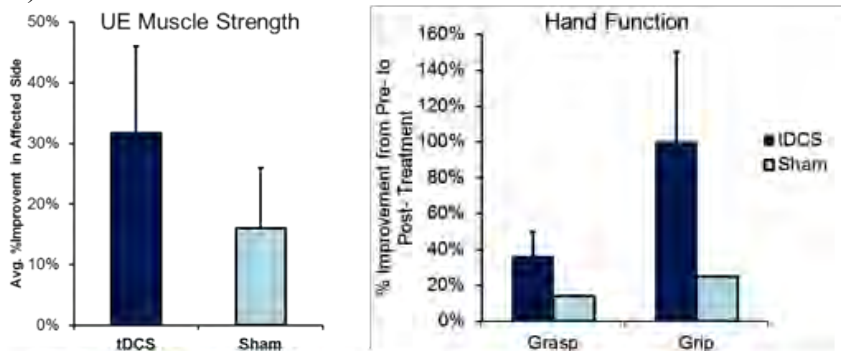
Ethnic/Racial Category	Sex/Gender		
	Females	Males	Total
Hispanic or Latino	0	0	0
American Indian/ Alaska Native	0	0	0
Asian	0	0	0
Native Hawaiian or Other Pacific Islander	0	0	0
African-American	0	1	1
White	0	10	10
Ethnic/Racial Category Total	0	11	11

D. Investigational Devices

N/A

E. Study Results

Patients in both groups have recovered in a short time frame of 2 weeks, but patients receiving tDCS plus rehabilitation have experienced more remarkable recovery. Muscle strength and function on grasp, grip and gross upper limb movements have improved. Patients in tDCS plus rehabilitation group show greater improvement in abilities of grasp and grip and a greater improvement in average muscle strength (Fig. 1B) as well as strength of weak proximal muscles as deltoid and biceps (Fig. 1C)



tDCS paired with therapy improves motor function: both manual muscle test scores in affected side in comparison to sham. In addition, the tDCS group demonstrated increased improvements in both grasp and grip in comparison to sham.

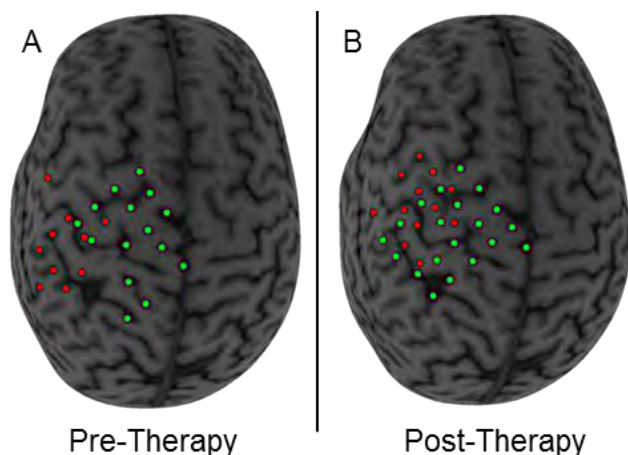


Fig 2. Loss of representation of weaker muscle (red) to that of spared muscles (green) after iSCI (A) is corrected in our study by pairing rehabilitation of weaker muscles with brain stimulation (tDCS) (B).

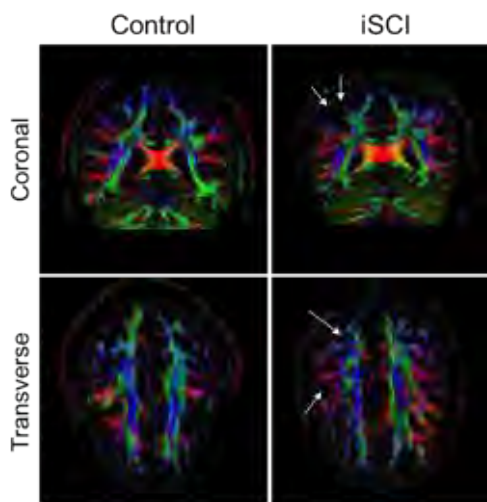
limb function than those receiving rehabilitation alone; these improvements are greater for more-affected muscles emphasized in training. In line with aim 2, it appears that these functional improvements in the more affected muscles may be explained by reorganization of their maps in the brain, which tend to enlarge and occupy M1 territory devoted to the less-affected muscle (Fig. 2A and B).

DTI imaging shows that changes in tracts emerging from motor cortex predict degree of recovery in our patients with SCI.

The improvement in tDCS group is paralleled by adaptive plasticity of maps of the more affected muscles that begin to share greater overlap with the less affected muscles.

Evaluation with TMS mapping shows maps in the brain devoted to more affected muscle and less affected muscle reorganize from pre- (Fig. 2A) to post-treatment (Fig. 2B). Maps devoted to more affected muscle, which was trained extensively, enlarge and show a shift towards map of the less-affected hand muscle.

Therefore, in line with aim 1 of the study, we have findings that indicate that patients with SCI receiving brain stimulation during rehabilitation show greater benefit in upper



Following iSCI, changes in cortical tracts occur (emphasized by arrows). Fiber direction is color coded in RGB. Red=right/left; green=anterior/posterior; blue=inferior-superior.

F. Summary of Anticipated and Unanticipated Adverse Effects

N/A

G. Deviations from Investigational Plan

No deviations from the investigational plan.

III. RISK ANALYSIS

A. New Adverse Information and New Risk Analysis

No new risks have been identified that would require a new risk analysis. The study is now registered as a pilot clinical trial. Use of tDCS, TMS and structural and functional MR imaging poses some risk, but strict adherence to inclusion-exclusion criteria, and protection against risks in conjunction with Clinical and Translational Science Association's Clinical Research Unit assistance has helped attenuate any safety threats.

IV. OTHER CHANGES

N/A

V. FUTURE PLANS

The study takes place at the Cleveland Clinic. Since Cleveland Clinic is not the primary site for clinical treatment for patients with SCI, recruitment was challenging at first. Still, our recruitment efforts have been successful, we have enrolled > 80% of the anticipated number of patients already and additional <20% patients will be recruited by end of Nov 2014. Since we are now benefitting from enhanced recruitment, we are seeking approval to complete study procedures on the already enrolled patients who are now completing therapies or are soon due to begin therapies. For instance, patients who start this month and next would require 1.5 months each to complete study therapies and then will be required to undergo one additional follow-up visit 3 months later. The reason for late start despite steady enrollment is that scheduling travel and lodging for a lot of these patients with paralysis following spinal cord injury poses the biggest challenge. Some of them are from out-of-town and can only manage studies in summer. We believe by extending the study by 1 more year, we would be able to complete recruitment goals and collect all data (including their 3-month follow-up visit) on all enrollees. This we believe would not impose any more costs on the budget than what remains for patients' care.

Therefore, since the study recruitment and data collection processes are proceeding well, we have requested approval to continue use of remaining funds through a no-cost extension period extension to simply collect all procedures on currently enrolled (>80% of anticipated) and due-to-be enrolled (<20% of anticipated) patients.

Annual Progress Report
For the period of 2013 through 2014

I. BASIC INFORMATION

SUBJECT: Expiratory Muscle Conditioning Using Functional Magnetic Stimulation (FMS) for Patients with Multiple Sclerosis

AWARD #: W81XWH-11-1-0707

CCF IRB#: 11-780 (expiration date: 9/29/2015)

SPONSOR: Department of Defense (DoD) Telemedicine and Advanced Technology Research Center

PRINCIPAL INVESTIGATOR: Vernon Lin MD PhD

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REPORT DATE: 11/07/2014

II. STUDY PROGRESS

A. Summary: Study Progress in Relation to Investigational Plan

The research study was reviewed and fully approved by Cleveland Clinic IRB. The expiration date is 9/29/2015. Xiaoming Zhang, PhD was hired in September 2012 as a co-investigator to manage the day-to-day activities of the project, participate in subject screening, data collection and analysis, manuscript and report preparation and dissemination of study results.

The MagPro R30 magnetic stimulator in July 2012 and the investigators completed training with the system. The team started to screen subjects with multiple sclerosis (MS) in September 2012. Inclusion criteria for this study are subjects with clinically defined MS whose baseline maximal expiratory pressure (MEP) values are between 50% and 70% of predicted. The medical records of approximately 200 MS patients were screened. The study has so far enrolled and completed data collection for 4 subjects.

B. Investigators/Investigational Sites

The study is being conducted at only one site – Cleveland Clinic Foundation. There are no plans to add another site.

Investigational Site	Investigators
Cleveland Clinic Foundation 9500 Euclid Avenue, Cleveland, OH 44195	Vernon Lin MD PhD Francois Bethoux MD Vinoth Ranganathan MSE MBA Xiaoming Zhang PhD Ela Plow PhD PT

All subject records and documentation will be kept in the FMS Laboratory at Cleveland Clinic Foundation.

C. Number of Subjects

Approximately 25 charts are screened each month to identify potential subjects. Four subjects have completed the protocol. The study has completed protocol for 3 subjects (two subjects in FMS arm and one subject in the RRMT arm) during the reporting period. One patient is still in the training protocol.

Ethnic/Racial Category	Sex/Gender		
	Females	Males	Total
Hispanic or Latino	0	0	0
American Indian/ Alaska Native	0	0	0
Asian	0	0	0
Native Hawaiian or Other Pacific Islander	0	0	0
African-American	0	0	0
White	2	2	4
Ethnic/Racial Category Total	2	2	4

D. Investigational Devices

N/A

E. Summary of Results

Three MS subjects with multiple sclerosis were recruited and completed the study protocol as of 10/31/14. Expiratory muscle activation was achieved by placing the center of the MC at T9 vertebral level. Baseline respiratory variables included maximal expiratory pressure (MEP), peak expiratory flow (PEF), and expiratory reserve volume (ERV). In this investigation, a six-week FMS conditioning protocol was performed. The stimulation intensity increased from 40% to 70%, progressively. The stimulation parameters were 20-Hz frequency, and 2-second stimulation duration. Pulmonary function tests were performed every two weeks. After 6 weeks of conditioning, the values for the main outcome measures (mean \pm standard error) were: MEP 77 ± 7.0 cmH₂O; PEF, 6.0 ± 2.2 L/sec; and ERV, 0.72 ± 0.15 L. These values corresponded to, 112%, 123%, and 120% of pre-FMS conditioning values, respectively. When FMS was discontinued for two weeks, these values had the following decrements (MEP, 3.5%, PEF, 15%, and ERV, 9.1%).

For the REMT group, the values for the main outcome measures, after 6-weeks of intervention, increased by 121% (MEP), 123% (PEF), and 120% (ERV) of pre-RRMT conditioning values, respectively. When REMT was discontinued for two weeks, these values had the following decrements (MEP, 3.5%, PEF, 9.4%, and ERV, 13%). A 6-week FMS conditioning of the expiratory muscles improved voluntary expiratory functions similar to traditional REMT, indicating that FMS may be a potential noninvasive therapeutic technology for training respiratory muscles in persons with multiple sclerosis.

FMS Group (N = 2)	ERV (L)	PEF (L/sec)	MEP (cmH₂O)
Baseline	0.6	4.86	69
2 weeks	0.67	4.91	77
4 weeks	0.68	5.66	77.5
6 weeks	0.72	6.01	77
post-test	0.62	5.21	74.75

REMT Group (N = 1)	ERV (L)	PEF (L/sec)	MEP (cmH₂O)
Baseline	0.23	2.57	66
2 weeks	0.2	2.3	77
4 weeks	0.17	3.05	77
6 weeks	0.31	3.74	80
post	0.27	3.39	78

F. Summary of Anticipated and Unanticipated Adverse Effects

N/A

G. Deviations from Investigational Plan

No deviations from the investigational plan.

III. RISK ANALYSIS

A. New Adverse Information and New Risk Analysis

No new risks have been identified that would require a new risk analysis.

B. Publications and Presentations

N/A

IV. OTHER CHANGES

N/A

V. FUTURE PLANS

The study team will continue to screen MS subjects for the study. Investigators will recruit 6 more subjects as specified in the study protocol. The enrollment rate has been very low because patients live outside of Cleveland or do not have transportation available to visit our research lab 5 days/week for 6-weeks. Study team expects to enroll the remaining study subjects in the next 6 months and complete the protocol.

Annual Progress Report
For the period of September 15, 2013 through September 30, 2014

I. BASIC INFORMATION

AWARD NUMBER: Proposal log number 10176004, Award Number W81XWH-11-10707

IRB NUMBER: 11-182 (expiration date: 04/07/2015)

STUDY NAME: A trial to compare the efficacy of Functional Magnetic Stimulation in enhancing GI motility in patients with constipation.

SPONSOR: Department of Defense (DoD) Telemedicine and Advance Technology Research Center

INDICATIONS FOR USE: Slow transit constipation

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REPORT DATE : November 7, 2014

II. STUDY PROGRESS

A. Summary: Study Progress in Relation to Investigational Plan

The study will evaluate the effects of functional magnetic stimulation (FMS) on colonic transit in non neurological constipated patients. Inclusion criteria for this study are subjects with clinically defined non-neurological constipation. Chronic functional constipation will be defined by the Rome II criteria and slow colonic transit will be documented by a Smart Pill which is wireless motility pill study. Eligible subjects will have a colonic transit time that is significantly longer than healthy subjects (>60hrs). The timing of treatment is a 5-week conditioning protocol. Sixteen patients are to be randomized to receive either the treatment or sham with a crossover design. Patients will be evaluated with the Smart Pill after receiving treatment. The site received IRB approval of the amendment (Protocol Version 3) on August 7, 2012 and approval from HRPO on 8/23/2012.

During the current reporting period, the study team was unable to recruit any subjects due to the following factors:

1. Patients do not want to be randomized as they need to set aside 10 weeks for treatment if they receive sham treatment. This is not feasible for working patients.
2. Smart Pill procedure prior to treatment was not budgeted for the study. It was to be billed to the patients insurance as it is considered standard of care. However, most insurance will not pay for it as they still continue to consider using SmartPill as investigational. As a result, most patients under going SmartPill procedure have to pay more than \$2000 as out of pocket expense which most patients cannot afford. Patients who pay this huge out-of pocket expense do not wish to be part of a study where they could be receiving sham treatment.
3. The enrollment rate has been difficult because most of the eligible patients live outside of Cleveland or do not have reliable transportation available to visit our research lab 5 days/week for 6-weeks.

Due to these issues, it has been very difficult to enroll subjects for this pilot study. The PI worked with HRPO to modify the protocol (remove the sham treatment). An amendment to this greater than minimal risk protocol was received by the HRPO on January 8, 2014. The amendment was approved by the Cleveland Clinic Institutional Review Board on December 13, 2013.

The amendment allows the following changes:

1. Reducing sample size from 16 to 8.
2. Removing the sham group.
3. Removing randomization.
4. Removing block design.
5. Cost of Smart Pills to be covered by institution.

The changes proposed in the amendment have been reviewed by the HRPO and found to be acceptable. The protocol amendment is approved (protocol version 4/dated 25 November 2013).

B. Investigators/Investigational Sites

The study is being conducted at only one site – Cleveland Clinic Foundation. There are no plans to add another site. No changes were made to the study team during this reporting period.

All regulatory documentations are kept at the Cleveland Clinic Foundation, 9500 Euclid Ave, Cleveland Oh 44195.

C. Number of Subjects

As of date, no patients have been enrolled or participated in the study. Since last approval, approximately 40 patients have been screened; 12 patients qualified; and 2 patients' insurance refused to cover the Smart Pill procedure. Unfortunately, after the amendment approval, neither of these two patients returned study coordinator's phone calls or emails updating and informing them that there was no out of pocket expense for the SmartPill.

D. Investigational Devices

N/A

E. Summary of Results

There are no results.

F. Summary of Anticipated and Unanticipated Adverse Effects

III. RISK ANALYSIS

A. New Adverse Information and New Risk Analysis

None

B. Publications and Presentations

None

IV. OTHER CHANGES

None

Investigational Site	Investigators
Cleveland Clinic Foundation, 9500 Euclid Avenue, Cleveland, Oh 44195	Massarat Zutshi,MD Xiaoming Zhang PhD Tracy Hull MD Brooke Gurland MD

V. FUTURE PLANS

We plan to enroll 8 patients. To inform and recruit more patients, our IRB has approved advertising study in social media websites: facebook, twitter, linkedin, and newsletter entitled, "Speaking of Women's Health" which is a program of the Cleveland Clinic Center for Specialized Women's Health <https://speakingofwomenshealth.com/column/read/controlling-your-bowels>. We have had numerous patients interested who have contacted us however they are not local and cannot remain in Cleveland for the required 5 weeks.

KEY RESEARCH AND ACCOMPLISHMENTS

Study 1: Magnetic stimulation and epilepsy

- The penicillin-induced seizure animal model has been generated by acute focal intracortical injection of penicillin in the motor cortex of rats.
- The effects of functional magnetic stimulation (FMS) on penicillin-induced seizure have been investigated using behavioral recording and electroencephalographic (EEG) recording.

Study 2: Directing Neuroplasticity to Improve Rehabilitative Outcomes of the Upper Limb in Incomplete Quadriplegia

- After treatments, patients with SCI receiving brain stimulation show greater benefit in upper limb function than those receiving rehabilitation alone.
- It appears that these functional improvements in the more affected muscles may be explained by reorganization of their maps in the brain, which tend to enlarge and occupy M1 territory devoted to the less-affected muscle.

Study 3: Expiratory Muscle Conditioning Using Functional Magnetic Stimulation for Patients with Multiple Sclerosis

- A 6-week FMS conditioning of the expiratory muscles improved voluntary expiratory functions, indicating that FMS may be a potential noninvasive therapeutic technology for training respiratory muscles in patients with Multiple Sclerosis.

Study 4: A prospective trial to study whether functional magnetic stimulation enhances gastrointestinal motility in patients with chronic constipation

- The PI worked with HRPO to modify the protocol (remove the sham treatment). The amendment was approved by the Cleveland Clinic Institutional Review Board on December 13, 2013.

REPORTABLE OUTCOMES:

Study 1: Magnetic stimulation and epilepsy

- Lin C-Y.**, K. Li, L. Franic, J. Gonzalez-Martinez, V.W. Lin, I. Najm, Y.-S. Lee. (2014). Frequency-dependent effects of contralateral repetitive transcranial magnetic stimulation on penicillin-induced seizures in rats. *Brain Research* **1581**: 103-116. PMID: 24937795.
- Lin C-Y.**, W. J. Huang, K. Li, R. Swanson, B. Cheung, V. W. Lin, Y.-S. Lee. (2014). Differential Intensity-dependent Effects of Magnetic Stimulation on Axons and Dendrites in Neuroscreen-1 Cells. *Journal of Neural Engineering* (submitted).
- Lin C.-Y.**, K. Li, V.W. Lin, I. Najm, Y.-S. Lee. (2015). Regulation of brain oscillation by contralateral repetitive transcranial magnetic stimulation on penicillin-induced seizures in rats. In preparation.

Study 2: Directing Neuroplasticity to Improve Rehabilitative Outcomes of the Upper Limb in Incomplete Quadriplegia

- Plow EB** and Fehlings MG (2014). [invited] Hypoxic locomotor rehabilitation for incomplete spinal cord injury: Not an oxymoron. *Neurology* 82(2): 98-99.
- Plow, E.B.**, Frost, F. S., Potter-Baker K, Chabra, P. “Testing brain stimulation’s potential to direct neuroplasticity and improve rehabilitation outcomes in spinal cord injury patients”. *Frontiers in Rehabilitation*

Study 3: Expiratory Muscle Conditioning Using Functional Magnetic Stimulation for Patients with Multiple Sclerosis

- Lin, VW** and Zhang, XM. “Evaluating Functional Magnetic Stimulation’s Potential to Improve Expiratory Function in Multiple Sclerosis Patients”. *Frontiers in Rehabilitation*

Conclusion

Study 1: Magnetic stimulation and epilepsy

The findings support the concept that MS increases BDNF expression and signaling, which sculpts axonal arborization and connectivity by which neuronal activity is regulated. Understanding the mechanism behind MS action is crucial in order to efficiently incorporate its use into potential therapeutic strategies.

Study 2: Directing Neuroplasticity to Improve Rehabilitative Outcomes of the Upper Limb in Incomplete Quadriplegia

The results indicate that patients with SCI receiving brain stimulation during rehabilitation show greater benefit in upper limb function than those receiving rehabilitation alone; these improvements are greater for more-affected muscles emphasized in training. It appears that these functional improvements in the more affected muscles may be explained by reorganization of their maps in the brain, which tend to enlarge and occupy M1 territory devoted to the less-affected muscle.

Study 3: Expiratory Muscle Conditioning Using Functional Magnetic Stimulation for Patients with Multiple Sclerosis

A 6-week FMS conditioning of the expiratory muscles improved voluntary expiratory functions, indicating that FMS may be a potential noninvasive therapeutic technology for training respiratory muscles in persons with MS; and continual FMS may be required to maintain gains in pulmonary functions.

Study 4: A prospective trial to study whether functional magnetic stimulation enhances gastrointestinal motility in patients with chronic constipation

During the current reporting period, the study team was unable to recruit any subjects. The PI worked with HRPO to modify the protocol (remove the sham treatment). An amendment to this greater than minimal risk protocol was received by the HRPO on January 8, 2014. The amendment was approved by the Cleveland Clinic Institutional Review Board on December 13, 2013.

References:

Study 1: Magnetic stimulation and epilepsy

- Li, K., Y.-S. Lee, L. Franic, J. Gonzalez-Martinez, V. W. Lin, I. Najm, **C.-Y. Lin**. The anti-convulsive effects of Transcranial Magnetic Stimulation (TMS) on penicillin-induced seizures. 2012, LRI retreat, Ohio (Corporate College East), USA.
- Li, K., Y.-S. Lee, L. Franic, J. Gonzalez-Martinez, V. W. Lin, I. Najm, **C.-Y. Lin**. The anti-convulsive effects of Transcranial Magnetic Stimulation (TMS) on penicillin-induced seizures. 2012, Cleveland Clinic Research Day, Ohio, USA.
- ***Lin, C.-Y.**, Y.-S. Lee, V. W. Lin, and J. Silver. (2012). Fibronectin inhibits chronic pain development after spinal cord injury. *Journal of Neurotrauma* **29**: 589-599. PMCID: PMC3278810 * corresponding author.
- Jiang, H.-H., O. N. Kokiko-Cochran, K. Li, **C.-Y., Lin**, V. W. Lin, M. Damaser, Y.-S. Lee. (2013). Bladder dysfunction changes from underactive to overactive after experimental traumatic brain injury. *Experimental Neurology*. **240**: 57-63. PMCID: PMC3552010
- Lee, Y.-S., K. Li, L. Franic, J. Gonzalez-Martinez, V. W. Lin, I. Najm, * **C.-Y. Lin**. 2013. Frequency-Dependent Effects of Transcranial Magnetic Stimulation on Penicillin-Induced Seizures in Rats. *Society for Neuroscience (submitted)*.
- Lee Y.-S., **C.-Y. Lin**, H.-H. Jiang, M. Depaul, M. Damaser, V. W. Lin, and J. Silver. (2013). Nerve regeneration restores supraspinal control of bladder function after complete spinal cord injury. *Journal of Neuroscience*. **33(26)**: 10591-10606. PMCID: PMC3693049
- Lee Y.-S., A. Danandeh, J. Baratta, **C.-Y. Lin**, J. Yu, and R.T. Robertson. (2013). Neurotrophic factors rescue basal forebrain cholinergic neurons and improve performance on a spatial learning test. *Experimental Neurology*. **249**: 178-186. PMCID: PMC3939719
- Park K.W., **C.-Y. Lin**, and Y.-S. Lee. (2014). Expression of Suppressor of Cytokine Signaling-3 (SOCS3) and its role in neuronal death after complete spinal cord injury. *Experimental Neurology* **261**: 65-75. NIHMS607759

Study 2: Directing Neuroplasticity to Improve Rehabilitative Outcomes of the Upper Limb in Incomplete Quadriplegia

B. Publications and Presentations

1. Cunningham DA, Machado A, Yue GH, Carey JR, **Plow EB** (2013). Functional somatotopy revealed across multiple cortical regions using a model of complex motor task. *Brain Research*. 1531:25-36. PMCID: PMC3931839.
2. **Plow EB**, Malone D, Machado A (2013). Deep brain stimulation of the ventral striatum/anterior limb of the internal capsule in thalamic pain syndrome: study protocol for a pilot randomized controlled trial. *Trials* 14:241. doi: 10.1186/1745-6215-14-241. PMCID: PMC3734205
3. **Plow EB**, Cunningham DA, Bonnett C, Gohar D, Bayram M, Wyant A, Varnerin N, Mamone B, Siemionow V, Hou J, Machado A, Yue GH (2013). Neurophysiologic Correlates of Aging-Related Muscle Weakness. *Journal of Neurophysiology* 110(11): 2563-2573. PMCID: PMC3882769
4. **Plow EB**, Cunningham DA, Beall E, Jones S, Wyant A, Bonnett C, Yue GH, Lowe M, Wang, X, Sakaie K, Machado, A (2013). Effectiveness and neural mechanisms associated with tDCS

- delivered to premotor cortex in stroke rehabilitation: study protocol for a randomized controlled trial. *Trials* 14: 331. PMCID: PMC3852558
5. **Plow EB** and Machado A (2013). Invasive Neuro-stimulation in Stroke Rehabilitation. *Neurotherapeutics* (2014) 11(3):572-82. PMCID: PMC4121447
 6. **Plow EB**, Varnerin N, Cunningham DA, Janini D, Bonnett C, Wyant A, Hou J, Siemionow V, Wang X, Machado AG, Yue GH (2014). Age-related Weakness of Proximal Muscle studied with Motor Cortical Mapping: A TMS Study. *PlosOne*. Feb 21;9(2):e89371. doi: 10.1371/journal.pone.0089371. PMCID: PMC3931763
 7. **Plow EB**, Cattaneo Z, Carlson TA, Alvarez GA, Pascual-Leone A, Battelli LB (2014). The compensatory dynamic of inter-hemispheric interactions in visuospatial attention revealed using rTMS and fMRI. *Frontiers in Human Neuroscience* Apr 17;8:226. doi: 10.3389/fnhum.2014.00226. PMCID: PMC4029023
 8. Machado AG, Gopalakrishnan R, **Plow EB**, Burgess RC, Mosher JC (2014). A Magneto-encephalography Study of Visual Processing of Pain Anticipation. *Journal of Neurophysiology*. Jul 15;112(2):276-86. PMCID: PMC4064417
 9. **Plow EB**, Cunningham DA, Varnerin N, Machado A (2014). Rethinking stimulation of brain in stroke rehabilitation: Why higher-motor areas might be better alternatives for patients with greater impairments. *Neuroscientist*. Jun 20. pii: 1073858414537381.
 10. Cooperrider J, Furmaga H, **Plow EB**, Park H-J, Chen Z, Kidd G, Baker K, Gale J, Machado A (2014). Chronic deep cerebellar stimulation promotes long-term potentiation, microstructural plasticity and reorganization of perilesional cortical representation in a rodent model. *Journal of Neuroscience*. 2;34 (27):9040-50. PMCID: PMC4078081.
 11. Cunningham DA, Machado A, Janini D, Varnerin N, Bonnett C, Yue GH, Jones SE, Lowe MJ, Beall E, Sakaie K, **Plow EB** (2014). Neural and Clinical Correlates of Interhemispheric Imbalance in Chronic Stroke. *Archives of Physical Medicine and Rehabilitation*. Sep 3. doi: 10.1016/j.apmr.2014.07.419
 12. Page SJ, Cunningham, DA, **Plow EB**, Blazak B. Neurorehabilitation and Non-Invasive Brain Stimulation: Two Great Tastes That Taste Great Together? *Archives of Physical Medicine and Rehabilitation (accepted)*